# GENE EXPRESSION OF PIP AQUAPORIN (VRPIP2;7) GENE IN VIGNA RADITA L. PLANT EXPOSURE TO MELATONIN UNDER DROUGHT STRESS CONDITIONS

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ABSTRACT: The study aimed to detect the *VrPIP2*;7 gene using PCR approach, as well as to know the effect of the treatment with four increased melatonin concentrations of 50, 100, 150 and 200 ppm in addition to control treatment were 0 ppm on the gene expression of plasma membrane intrinsic proteins (PIP) genes in *Vigna radiata* L. plant exhibition for five periods of drought which is irrigation every 24 hours, 48 hours, 5 days, 10 days and every 15 days.

The electrophoresis of agarose gel at a concentration of 2% showed one band when detecting the *VrPIP2*;7 gene with a sizeable 732 bp and using the 100 bp volume index. This gene was selected for sequencing study based on its importance as well as on the results of its gene expression. The sequencing of this gene was recorded on the global site, the National Center for Biotechnology Information (NCBI) and was given an accession number for nucleotide sequences obtained with MT992077.1. The gene expression of the plasma membrane aquaporin gene was studied at the roots of the melatonin-prone mung bean plant under the influence of drought and the results showed that the gene expression of *VrPIP2*;7 gene was good and gave remarkable results and for all the periods used, as it was observed at the irrigation period every 24 days and when the concentration of melatonin increased from zero ppm to 100 ppm gene expression increased from 1 to 1.11, as well as the concentration of 150 ppm, gene expression increased to 1.38 compared to control plants, while the irrigation period every 48 hours was observed that all concentrations caused an increase in gene expression with a concentration of more than 50 ppm. The results showed that the irrigation period every 5 days was also good, with an increase in the expression of the *VrPIP2*;7 gene and for most of the concentration used with a concentration of more than 50 ppm by giving it the highest genetic expression and an increase of 43% compared to control plants.

The results also indicated that at the irrigation period every 10 days, increased concentrations of melatonin led to an increase in the expression of the *VrPIP2;7* gene, and it was noted that concentrations of 50 and 200 ppm had increased gene expression with a concentration of more than 200 ppm compared to control plants. The results showed that the irrigation period every 15 days had an effect on the increase in the gene expression of *VrPIP2;7* gene and for all the increased concentrations of melatonin with a concentration of more than 150 ppm compared to control plants.

Key words: Vigna radiata L., VrPIP2;7, PIP genes, drought stress.

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### INTRODUCTION

Vigna radiata L. (Mung bean) is one of the plants belonging to the Leguminosae family (Author, 1988) and it is an annual herbaceous summer crop. Its stem is standing or semi-upright branched and covered with fluff; its height ranges between 25-125 cm, and contains triple compound leaves. The flowers of mung bean are with yellowish to yellowish-green color grouped in panicle inflorescences consisting of 5-15 flowers; its horny fruits

(leguminous) covered with fluff with green to brown color. The cultivation of mung is spread in most countries of the world, but the tropical and semi-tropical regions are more cultivated, especially in India, China, Bangladesh and Thailand, and it is characterized by being one of the most producing countries of this crop; the cultivation rate of mung is 4 million hectares annually in South and East Asia in the world out of the total cultivation in the world of 5.8 million hectares annually (Al-Janabi and Ali, 1996;

from 1 to 1.92, with rate of 92% compared to the control plants. Similar result was found after using the concentration 150 ppm and the gene expression increased from 1 to 1.95, with rate of 95% in comparison to the control plants, while the concentration 200 ppm increased the gene expression from 1 to 1.40, with rate of 40% compared to the control plants.

The reason for this may be due to the fact that it is known that water stress reduces the hydraulic conductivity of the roots and thus inhibits the growth of roots and plants (Azaizeh et al, 1992; Carvajal et al, 1999) by decreasing the level of aquaporins, mainly PIPs and TI'Ps (Bursiac et al, 2005), in addition to its association with dynamic changes in post-translational modifications such as phosphorylation and mediation that affect the function of aquaporins (Dipietro et al, 2013). Therefore, the high levels of VrPIP2;7 gene expression will molecularly overcome the decrease in the levels of endogenous aquaporins during these stresses, leading to higher water flow to the roots and leaves, which in turn helps to grow under light to moderate stress. This gene is may responsible for increasing of root length (Khan et al, 2015) and this result is in agreement with Khan et al (2015) study on Arabidopis thaliana. Moreover, the activity of occopurines decreases under drought stress due to an imbalance in the regulation of hydraulic conductivity of the roots (Qiao et al, 2020). Interestingly, the exogenous melatonin treatment may regulate the gene expression of PIPs and improve the hydraulic regulation of the roots, which caused an increasing their water absorption capacity (Tiwari et al, 2020) and these findings are in agreement with Qiao et al (2020) study on the corn plant (Zea mays).

#### REFERENCES

- Abdul Malik A (2015) Analysis of the resistance of durum wheat (*Triticum turgidum* var *durum* L.) to abiotic stresses in the late growth phase. *PhD thesis*, College of natural and biological sciences, Farhat Abbas University, Setif, Ministry of Higher Education and Scientific Research, Republic of Algeria, 221 pp.
- Agre P, Sasaki S and Chrispeels M J (1993) Aquaporins: a family of water channel proteins. Am. J. Physiol. - Renal Physiol. 265, F461-F461.
- Ali M Ch, Talib E and Jadaan H M (1990) *Legume crops*. Al-Hikma house for printing and publishing. Baghdad, Iraq. pp: 58-68.
- Al-Kateb Y M (1988) Classification of seed plants. University of Baghdad, Ministry of Higher Education and Scientific Research.
- Azaizeh H, Gunse B and Steudle E (1992) Effects of NaCl and CaCl<sub>2</sub> on water transport across root cells of maize (*Zea mays* L.) seedlings. *Plant Physiology* **99**, 886–894. pmid:16669016.
- Bashar A S (2013) An economic analysis of the response to strawberry disease in Iraq for the period from 1970-2010. *Iraqi J. Agricult. Sci.* **44**(2), 258-263.

- Boursiac Y, Chen S, Luu D T, Sorieul M, van den Dries N and Maurel C (2005) Early effects of salinity on water transport in Arabidopsis roots. Molecular and cellular features of aquaporin expression. *Plant Physiol.* **139**, 790–805.
- Bowler C, Van Montagu M and Inzé D (1992) Superoxide dismutases and stress tolerance. *Ann. Rev. Plant Physiol. Plant Mol. Biol.* **43**, 83-116.
- Carvajal M, Martinez V and Alcaraz C F (1999) Physiological function of water channels as affected by salinity in roots of paprika pepper. *Physiologia Plantarum* **105**, 95–101.
- Dat J, Vandenabeele S, Vranová E, Van Montagu M, Inzé D and Van Breusegem F (2000) Dual action of the active oxygen species during plant stress responses. *Cell. Mol. Life Sci.* **57**, 779-795.
- Di Pietro M, Vialaret J, Li GW, Hem S, Prado K and Rossignol M (2013) Coordinated post-translational responses of aquaporins to abiotic and nutritional stimuli in Arabidopsis roots. *Mol Cell Proteomics* **12**, 3886–3897 doi: 10.1074/mcp.M113.028241.
- Fischer T W, Kleszczynski K, Hardkop L H, Kruse N and Zillikens D (2013) Melatonin enhances antioxidative enzyme gene expression (CAT, GPX, SOD) prevents their UVR induced depletion and protects against the formation if DNA damage (8-hydroxy-2deoxygaunosine) *in* sex *vivo* human skin. *J. Pineal. Res.* **54**, 303-312.
- Javot H, Lauvergeat V, Santoni V, Martin-Laurent F, Guclu J, Vinh J, Heyes J, Franck K I, Schäffner A R, Bouchez D and Maurel C (2003) Role of a single aquaporin isoform in root water uptake. Plant Cell. 15, 509-522.
- Kaldenhoff R and Fisher M (2006) Aquaporins in plants. *Acta Physiol.* **187**, 169–176.
- Khan K, Agarwal P, Shanware A and Sane V A (2015) Heterologous Expression of Two Jatropha Aquaporins imparts drought and salt tolerance and improves seed viability in transgenic *Arabidopsis thaliana*. *PLoS One* **10**(6), e0128866. doi:10.1371/journal.pone.0128866.
- Kole C (2011) Wild crop relatives: Genomic and breeding resources. Vol.: Legume Crops and Forages. Springer Berlin, Heidelberg, Germany, pp.1-19.
- Maurel C, Tancet F, Guclu J, Guern J and Ripoche P (1997) Purified vesicles of tobacco cell and vacuolar membranes exhibit dramatically different water permeability and water channel activity. *Proceed. Nat. Acad. Sci. USA* **94**, 7103-7108.
- Mittler R (2002) Oxidative stress antioxidants and stress tolerance. *Trends Plant. Sci.* **7**, 405-410.
- Qiao Y, Ren J, Yan Y, Liu Y, Deng X, Liu P and Wang Sh (2020) Exogenous melatonin alleviates PEG- induced short-term water deficiency in maize by increasing hydraulic conductance. *J. BMC Plant Biol.* **20** (218), 10 .1186/s12870-020-02432-1.
- Rafiq M, Mali M, Ahmad Naqvi S H, Umar Dahot M, Faiza H and Khatari A (2012) Regeneration of plants in EMS treated local mungbean under salt stress. *Pak. J. Biotechnol.* **9**(2), 83-89.
- Shekoofe A and Sinclair Th (2018) Aquaporin activity to improve crop drought tolerance. *Cells* **7**(123), 1-10.
- Tan D X, Hardeland R, Manchester L C, Reiter R J, Plummer B, Limson J, Weintraub S and Qi W (2000) Melatonin directly scavenges hydrogen peroxide: apotentially new metabolic pathway of melatonin biotransformation. *Free Radical. Biot & Med.* 29, 1177-1185.
- Touati M (2002) The effect of two water stress methods on osmoyic

- adjustment solute accumulation and expensive drought in two durum wheat varieties (*Triticum durum* Desf). These de magistere. ENS. Kollba. Alger., 115 p.
- Trillo N and Fernandez R J (2005) Wheat plant hydraulic properties under prolonged experimental drought: Stronger decline to root system conductivity than in leaf area. *Plant Soil* **277**, 277–284.
- Vranova E, Inzé D and Van Breusegem F (2002) Signal transduction during oxidative stress. *J. Exp. Bot.* **53**, 1227-1236.
- Willekens H, Chamnongpol S, Davey M, Schraudner M, Langebartels C, Van Montagu M, Inzé D and Van Camp W (1997) Catalase is

- a sink for  $H_2O_2$  and is indispensable for stress defence in C3 plants. *Europ. Mol. Biol. Org. J.* **10**, 1723-1732.
- Yang S, Zhang X, Xu Y and Zhou X (2002) Rapid determination of serum melatonin by ESI-MS-MS with direct sample injection. *J. Pharm. Biomed. Anal.* **30**, 781-790.
- Zhang N, Zhao B, Zhang H J, Weeda S, Yang C, Yang Z C, Ren S X and Guo Y D C (2013) Melatonin promotes water stress tolerance, lateral root formation and seed germination in cucumber (*Cucum issativus* L.). *J. Pineal. Res.* **54**, 15-23.